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PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of)
••) Group Art Unit: 1645
ZHELNIN and BLOOMQUIST)
) Examiner: S. Gucker
Serial No. 09/899,532)
)
Filing Date: July 6, 2001) Docket No. 02973.00040

For: HUMAN NEUROPEPTIDE Y-LIKE G PROTEIN-COUPLED RECEPTOR

SUBMISSION OF SUBSTITUTE DECLARATION UNDER 37 C.F.R. § 1.131

U.S. Patent and Trademark Office 220 20th Street S. Customer Window, Mail Stop Amendment Crystal Plaza Two, Lobby, Room 1B03 Arlington, VA 22202

Dear Sir:

Applicants responded on June 25, 2004 to the Office Action mailed February 25, 2004 in the application referenced above. The response included a declaration of the inventors under 37 C.F.R. § 1.131, which included Exhibits 1-10. The declaration stated that the dates on Exhibits 1-10 had been redacted; however, the Exhibits 1-10 filed together with the declaration inadvertently contained dates.

A substitute declaration with Exhibits 1-10 accompanies this paper. The declaration itself is a copy of the one filed June 25, 2004. Exhibits 1-10 are identical to those filed June 25, 2004 except that the dates have been redacted.

Please <u>substitute</u> the declaration and exhibits that accompany this paper for those filed June 25, 2004. Please discard the declaration and exhibits filed June 25, 2004 and delete all copies of the declaration and exhibits that may have been scanned electronically.

We believe no fee is associated with this request. If a fee is required, please charge our Deposit Account No. 19-0733.

Respectfully submitted,

BANNER & WITCOFF, LTD.

Date: July 6, 2004

By:

Lisa M. Hemmendinger Registration No. 42,653

Customer No. 22907





IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:)	
ZHELNIN and BLOOMQUIST)	Group Art Unit: 1645
Serial No. 09/899,532))	Examiner: S. Gucker
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For: HUMAN NEUROPEPTIDE Y-LIKE G PROTEIN-COUPLED RECEPT)R

DECLARATION UNDER 37 C.F.R. § 1.131

U.S. Patent and Trademark Office 220 20th Street S. Customer Window, Mail Stop Non-Fee Amendment Crystal Plaza Two, Lobby, Room 1B03 Arlington, VA 22202

Dear Sir:

We, Leonid Zhelnin and Brian Bloomquist, declare as follows:

- 1. We are the named inventors of the subject matter claimed in the pplication referenced above.
- 2. Prior to March 2, 2000, we reduced to practice the subject matter of claims 1-8 and 12 of the application referenced above. The dates on the attached exhibits have been reducted; however, all the work described in this declaration was performed in the U1 ited States prior to March 2, 2000.
- 3. Prior to March 2, 2000, we monitored search results from a prog im (Gene Agent) that automatically checked sequences released into the public domain for he nology to the orexin and NPY1 receptors. Prior to March 2, 2000, we received reports from the orexin-

and NPY1-receptor Gene Agent searches, respectively, indicating that DNAs with 1 accession numbers EM:AC005961 (Homo sapiens Chromosome 11q12.2 PAC pDJ32L16 gen mic DNA) and GB:g3823006 (Human sperm genomic library) both had homology to the crexit and NPY1 receptor sequences. Copies of the reports are attached as Exhibit 1. Sequence analy is revealed these DNAs to be overlapping and that AC00596 contained 154,647 nucleotides comprising what appeared to be at least five exons with homology to the crexin receptor. A copy of BLAST comparisons of the translated five exons with the amino acid sequence of the crexir receptor is attached as Exhibit 2.

- 4. Prior to March 2, 2000, we used sequence information from AC00596: to amplify by polymerase chain reaction (PCR) amplification two PCR products of 234 and 1 1 bp from genomic DNA. The longer 234 bp clone included both transmembrane domains (TM 1 and 2 of the putative GPCR. A copy of the nucleotide sequence of the PCR product is attached as Exhibit 3.
- 5. We designed primers to the 5'- and 3'-regions of the 234 bp clone, and 5'- and 3'-RACE amplifications were carried out to clone the full coding region of the putative < PCR. The reverse primers used in the strategy were termed "6A1-85B" and "6A1-126B" and an shown on pages 2 and 3 of Leonid Zhelnin's laboratory notebook no. RB53651. Copies are 1 rovided as Exhibit 4.
- 6. Prior to March 2, 2000, we obtained two 5'-RACE clones (~220 and ~320 bp) from human heart cDNA; sequence analysis of the 5'-RACE clone 5GA1 revealed that it encodes a peptide with homology to the N-terminal 105 amino acids of NPY1. A copy of age 47 of Leonid Zhelnin's laboratory notebook no. RB53651 showing a ClustalW-formatted al gnment of the amino acid sequence encoded by 5GA1 and the amino acid sequence of the neurop ptide Y-1

receptor (NPY-1R.pep) is provided as Exhibit 5. The position of the putat /e START methionine codon (ATG) in clone 5GA1 was consistent with clone 5GA1 being si nilar to the NPY-1 and orexin-2 receptors. The identity of this ATG as the true START codon was unconfirmed, however, because the open reading frame (ORF) was not closed upstres n of the 5'-most ATG.

- 7. Sequence analysis of the 3'-RACE amplicon (~0.9 kb in length; 3 -RACE #2 clone) revealed that this clone contained sequences homologous to a GPCR through IM6. The primers used in the 3'-RACE strategy were termed "6A1-F32" and "6A1-F41" and at : shown on pages 4 and 5 of Leonid Zhelnin's laboratory notebook no. RB53651. Copies are rovided as Exhibit 6. However, the 3'-RACE amplicon lacked both a TM7 domain and corresponding STOP codon. A complete clone was electronically assembled from the 3'-RACE amplicon and clone 5GA1 to create a 1047-bp sequence contig, which encoded 349 amino acids and which was named GA1. A copy of page 53 of Leonid Zhelnin's laboratory notebook no. RB536 1 showing the electronically assembled sequence is provided as Exhibit 7.
- 8. Prior to March 2, 2000, GA1 was used as template to search he Image Consortium database for novel expressed sequence tags (ESTs, which are cDNAs) with identity to GA1 to identify the missing 3'-end. Three Image clones from human kidney were identified and found to have similarity based on our previous sequence analysis. Identification of the three Image clones was recorded on page 61 of Leonid Zhelnin's laboratory notebook no. F B53651; a copy is provided as Exhibit 8.
- 9. Sequence analysis of Image clone 2055185 confirmed the putative START methionine because sequence analysis revealed it to contain an in-frame STOP codo: upstream

of the putative START methionine. This sequence information is summarized or page 89 of Leonid Zhelnin's laboratory notebook no. RB53651; a copy is provided as Exhibit 9.

- 10. Under our direction the 3'-ends of the Image cDNAs (from the NCI_CGAP_Kid12 and Soares NFL_T_GBC_S1 libraries) were sequenced. One c the Image clones had an extended 3'-end that included the presence of a 710-bp intron folloved by what appeared to be further exonic sequence.
- 11. The sequence immediately downstream of the intron of this Image c NA clone was used as the query to search (via the BLAST algorithm) the genomic sequence within AC005961 to identify the missing 3'-coding region. Prior to March 2, 2000, BLA 'T analysis identified the missing 271 nucleotides of the 3'-end of the coding region of the G 'CR. This sequence information is summarized on page 89 of Leonid Zhelnin's laboratory no tebook no. RB53651. See Exhibit 9.
- 12. Prior to March 2, 2000, we amplified clones containing the comple e 1293-bp ORF from both human heart and brain cDNA by PCR (using Marathon-Ready cDN 1 libraries, Clontech) and recorded this information on page 89 of Leonid Zhelnin's laboratory no tebook no. RB53651. See Exhibit 9.
- 13. Also prior to March 2, 2000, the coding sequence of the full-length cl 'NA clone was confirmed by sequence analysis of a full-length amplicon generated by PCR fi on human brain cDNA. This sequence was recorded on pages 95-97 of Leonid Zhelnin's laboratory notebook no. RB53651; copies of these pages are provided as Exhibit 10. The coding sequence is contained within SEQ ID NO:1 of our provisional application Serial No. 60/215,523 and within SEQ ID NO:1 of our utility application 09/899,532. This coding sequence e codes the 431 amino acid sequence shown in SEQ ID NO:2 of both the provisional and utility applications.

14. All statements made herein of our own knowledge are true and all statements made on information and belief are believed to be true; and further that these state nents were made with the knowledge that willful false statements and the like so made are pullishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any palent issuing thereon.

Dated: June 25, 2004

Brian Bloomquist, Ph.D.

Dated: June 25, 2004

Leonid Zhelnin, Ph.D.



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7.40-17

Analysis Results

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Requested by: Ebelmin, Leonid on

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Project Summary

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Requested by: Zhelmin, Leonid

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INCYTE PHARMACBUTICALS, INC.

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Score = 103 (48.3 bits), Expect = 1.8e-12, Sum P(5) = 1.8e-12 Identities = 17/52 (32%), Positives = 33/52 (63%), Frame = (+2)

Query: 112 LVVDITETWFFGQSLCKVIPYLQTVSVSVSVLTLSCIALDRWYAICHPLMFK 163
L+ D++ G +CK++P++Q+ +V +LT++CIA++R + HP K

Sbjct: 48452 LMYDLSYFLTAGAFICKMVPFVQSTAVVTEILIMTCIAVERHQGLVHPFKMK 48607

Score = 88 (41.3 bits), Expect = 1.8e-12, Sum P(5) = 1.8e-12 Identities = 15/54 (27%), Positives = 31/54 (57%), Frame = (+3)

Query: 210 CDERWGGEIYPKMYHICFFLVTYMAPLCLMVLAYLQIFRKLWCRQIPGTSSVVQ 263

C E W ++ K+Y ++ ++ PL +M++ Y +1 +LW ++ G SV++

Sb_ct: 56082 CLBEWTSPVHQKIYTTFILVILPLLPLMVMLILYSKIGYELWIKKRVGDGSVLR 56243

Score = 71 (33.3 bits), Expect = 1.8e-12, Sum P(5) = 1.8e-12 Identities = 15/54 (27%), Positives = 26/54 (48%), Frame = (+1)

Query: 329 VFGMFAHTEDRETVYAWFTFSHWLVYANSAANPIIYNFLSGKFREEFKAAFSCC 382 + G F D T+ F + ++NS NPI+Y F++ F++ +A C Sbjct: 59380 ISGNFEKEYDDVTIKMIFAIVQIIGFSNSICNPIVYAFMNENFKKNVLSAVCYC 59541

Score = 47 (22.1 bits), Expect = 1.8e-12, Sum P(5) = 1.8e-12 Identities = 9/17 (52%), Positives = 11/17 (64%), Frame = (+2)

Query: 46 EYLHPKEYEWVLIAGYI 62 EY H +E W LI+ YI

Sbjct: 29786 EYPHAEE*NWTLISQYI 29835

Score = 46 (21.6 bits), Expect = 1.8e-12, Sum P(5) = 1.8e-12 Identities = 6/8 (75%), Positives = 7/8 (87%), Frame = (+3)

Query: 381 CCCLGVHH 388

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Sbjct: 148295 CCCLGLEH 148319



Retrieve FASTA Sequences

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Created By: Zhelnin, Leonid

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• 10 mM Tricine-KOH (pH 9.2)	cloning. Enough material is provided for 30	26
• 1 mM EDTA	50-μi Marathon RACE reactions.	27
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 -20°C Avoid multiple freeze/thaw cycles. 	POLY A' RNA SOURCE:	29
	Normal, whole hearts pooled from 3 male	30
SHELF LIFE: I year from date of receipt under	Caucasians, ages 28-47; cause of death: trauma	11
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CROSS REFERENCES

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